# MORPHOLOGY, CHROMOSOME NUMBER, AND FLAVONOID CHEMISTRY OF BIDENS CORDYLOCARPA (COMPOSITAE)

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Coreopsis cordylocarpa was described by Gray in 1887. For many years no one seems to have questioned that this species had an appropriate generic assignment. In his two comprehensive treatments of *Coreopsis*, Sherff (1936, 1955) does not even suggest that *C. cordylocarpa* might have been improperly placed. In his later years, however, he indicated a specimen in the Field Museum (*Cronquist 9779*) as the type of a new *Bidens* species which he proposed to name in honor of Arthur Cronquist. The name was never published, and Sherff later recognized the Cronquist specimen as belonging to *Coreopsis cordylocarpa*, and so annotated it.

My first encounter with this species in the field was in the state of Jalisco in the late summer of 1966, where I was collecting with T. Melchert and P. Sorensen. All three of us were somewhat familiar with members of the Coreopsidinae, and it seems significant that our first impression was that we were observing a species of *Bidens*. In view of this, along with the fact that Sherff had also at one time assigned this taxon to *Bidens*, a detailed study seemed indicated. The present paper gives the result of this study.

## MATERIALS AND METHODS

For the cytological studies, floral buds were fixed in the field in chloroform: absolute ethyl alcohol: glacial acetic acid (4:3:1). The anthers were squashed in aceto-hematoxylin, and the chromosomes observed in dividing microsporocytes.

Leaves and flowers which were collected in the field and dried, served as one source of flavonoids for chromatographic analysis. Achenes were collected at the same time, and fresh material from plants grown in the greenhouse was also analyzed for flavonoid constituents. The floral tissues (ray floret corollas, disk floret corollas and adnate anthers, disk floret ovaries, and chaff) were analyzed separately and found to be chromatographically identical. The leaf profiles were also determined. The tissues were placed in 0.1% HCl in methanol for 24-48 hours. This extract was applied to Whatman 3MM chromatographic paper ( $46 \times 57$ cm sheets) and run in two dimensions by the descending method. The first solvent system was tertiary butyl alcohol: glacial acetic acid: distilled water (3:1:1 v/v); the second glacial acetic acid: distilled water (15:85 v/v). Drawings of these chromatograms are shown in Fig 5. In these figures each chalcone-aurone pair is designated by a single letter and represented as a single spot because they invariably occur together as a complex mixture.

Individual flavonoids were purified by repeated chromatography, and

entity etin-3- coside genin-7- coside psin	MeOH 359 295 265a 255 330 283	+A1C1 <sub>3</sub> 432 330 300a 274 385 306	+A1C1 <sub>3/</sub> HC1 404 365 300a 267 385 306	+NaOMe 413 330 272 445 390a 286		381 295a 261 330 283
coside genin-7- coside	295 265a 255 330 283	330 300a 274 385	365 300a 267 385	330 272 445 390a	325 271 330	295a 261 330
coside genin-7- coside	295 265a 255 330 283	330 300a 274 385	365 300a 267 385	330 272 445 390a	325 271 330	295a 261 330
genin-7- coside	265a 255 330 283	300a 274 385	300a 267 385	272 445 390a	271 330	261 330
coside	255 330 283	274 385	267 385	445 390a	330	330
coside	330 283	385	385	390a		
coside	283			390a		
		306	306		283	283
psin	383			286		
psin	383					
psin	383					
psin	383					
		505a	440	450	480a	510
	295	450	320	380a	385	413
	260	318	272	285	288	345
	245	275	250		255a	285
		250				
rein	403	450	405	488	487	435
	340a	342	335a	345	405	337
	274	292	275	291	340a	288
	256	255a	257		278	259
					255	
in	382	520a	420	452	383	393
	320a	429	332	340	325a	320
	263	332	272	285	265	270
		273	250	253		
imein	416	455	413	497	443	442
	330	328	325	348	365	328
	273	287	273	290a	259	282
	242	248a	243	261		245
maturer						
	n-sulfure mixture	330 273 242 n-sulfure mixture?	330 328 273 287 242 248a n-sulfure	330     328     325       273     287     273       242     248a     243       n-sulfure-	330     328     325     348       273     287     273     290a       242     248a     243     261	330   328   325   348   365     273   287   273   290a   259     242   248a   243   261

TABLE	1	
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IDENTIFICATION AND SPECIAL MAXIMA OF THE FLAVONOIDS OF C. CORDYLOCARPA

the compounds were finally analyzed spectrally, utilizing a Beckman DB-G Grating Spectrophotometer. Standard methods and diagnostic reagents were employed (Markham and Mabry, 1968; Jurd, 1962). The spectral properties of these compounds, together with their identifications, are given in Table 1.

### RESULTS AND DISCUSSION

There are a combination of morphological features which serve to distinguish this species from other taxa in the genus *Coreopsis* found in



FIG. 1. Distribution of C. cordylocarpa.

Mexico. The (8)10-16 ray florets, club-shaped wingless achenes (fig. 3B), fruticose habit, relatively undifferentiated outer and inner involucral bracts (fig. 2B), and large (to 20 cm) pinnatisect, deltoid leaves (fig. 4A) are quite unique. Sherff (1955) treated *C. cordylocarpa* as a member of sect. *Coreopsis*. It is clearly a discordant element here, however, for all other species in this section are small annual or perennial herbs with dorsiventrally flattened, winged achenes. In fact, all species of *Coreopsis* which I have examined (primarily those from North America) have achenes which are variously flattened dorsiventrally. That Sherff placed *C. cordylocarpa* in the type section seems to indicate a lack of understanding of its affinities within the genus *Coreopsis*.

Certain morphological features of C. cordylocarpa are much more suggestive of some Mexican Bidens species than they are of any member of Coreopsis. Specifically, the outer and inner involucral bracts of C. cordylocarpa are quite similar in shape (fig. 2B), and differ primarily in color, the outer ones being dark green, whereas the inner are pale green to nearly white. These involucral characteristics are very similar to those encountered in many species of Mexican Bidens. In contrast, species of Mexican Coreopsis typically exhibit a highly dimorphic involucre with somewhat green and fleshy outer bracts which differ from the inner ones in shape, size, color, and texture (fig. 2A). Moreover, the elongate, club-shaped, terete, striate, and wingless achenes of C. cordylocarpa are similar in general shape and appearance to those of several species of Bidens from Mexico (fig. 3A, B). Certainly, the achenes of C. cordylocarpa in no way resemble the flat, winged fruits which are typical of all Mexican Coreopsis, and indeed of the genus as a whole (fig. 3B, C).

The chromosome number of C. cordylocarpa offers no clues as to its

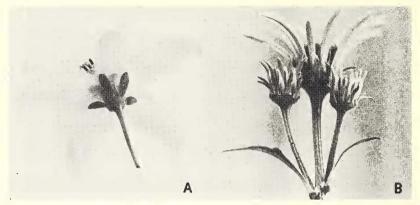
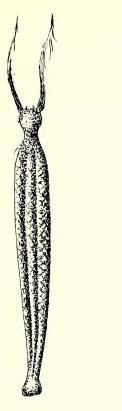


FIG. 2. Photographs of filoral heads. A, floral head of typical Mexican Coreopsis showing the highly dimorphic involucre; B, floral head of C. cordylocarpa showing the undifferentiated outer and inner involucral bracts.

generic affinities. A count of  $2n = 146 \pm 2$  (fig. 4B) was determined in a large number of cells from *Melchert*, *Sorensen*, & *Crawford* 6347A. It must be emphasized that observations from several other populations (*Melchert*, *Sorensen*, & *Crawford* 6354 & 6371; *Carman* 68-60) revealed a chromosome complement of  $2n = 146 \pm 6-8$ . From these data, it appears justifiable to conclude that only one ploidy level exists in *C. cordylocarpa*, and that probably all populations have the same or nearly the same chromosome number. This high number, unique in the Coreopsidinae and one of the highest reported in the Compositae, is particularly interesting from an evolutionary point of view when considered together with the geographic distribution and ecology of the species. *Coreopsis cordylocarpa* is endemic to Jalisco, Mexico (fig. 1) and occurs only in or along the banks of shallow streams, indicating that it may be an old species, representing the only extant taxon of an otherwise extinct polyploid complex.

The flavonoid chemistry of *C. cordylocarpa* suggests a closer affinity to other Mexican species of *Bidens* than to any *Coreopsis* taxon. As shown in Fig. 5, the leaves and floral tissues are dominated by two chalcone-aurone pairs. Coreopsin-sulfurein (spot A) and marein-maritimein (spot F) are invariably present in large quantities in both leaves and flowers. In addition, the leaves (fig. 5, left) contain a flavonol (spot 6, quercetin-3-glycoside) and a flavanone (spot 16, naringenin-7glycoside). The flowers also contain spot X (fig. 5, right), which appears to be a mixture of butein and sulfuretin, these being the aglycones of coreopsin and sulfurein respectively.

Chemical analysis of the leaves of other suffruticose or fruticose *Coreopsis* species from Mexico (members of sections *Electra*, *Anathysana*, and *Pseudo-Agarista*) has revealed the complete absence of coreopsin-sulfurein and marein-maritimein. These compounds are sometimes present in the floral tissues of certain of these species, but never



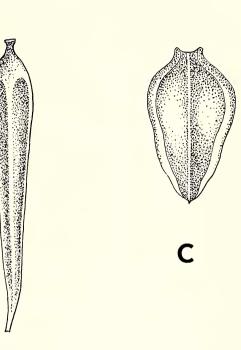


FIG. 3. Drawing of *Coreopsis* and *Bidens* achenes. A, achene of a typical Mexican *Bidens*; B, achene of *C. cordylocarpa*; C, achene of a typical Mexican *Coreopsis* (all  $\times$  ca. 5).

in the leaves. It must be admitted that sufficient data are not available to make a meaningful statement concerning the distribution of these substances in the genus *Coreopsis* as a whole. However, it is instructive to compare the leaf profile of *C. cordylocarpa* to those of several species of *Bidens* from the United States and Mexico. The leaves of these taxa contain an unidentified chalcone-aurone pair which is chromatographically very similar (probably identical) to marein-maritimein. This evaluation is based upon conversations with T. E. Melchert and my observations of numerous chromatograms of the leaves of *Bidens* species. Although the chemical evidence is not conclusive, it certainly suggests that *C. corylocarpa* is much more similar to *Bidens* in its flavonoid chemistry than it is to *Coreopsis*.

Since the general morphology, as well as the preliminary chemical data, suggest that the affinities of *C. cordylocarpa* are with *Bidens* rather than with *Coreopsis*, the following new combination is proposed.

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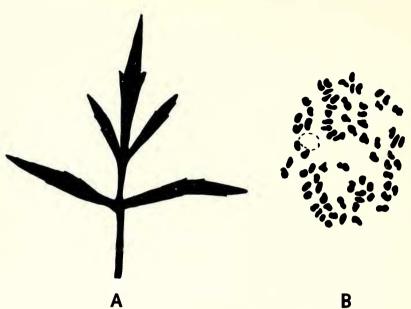


FIG. 4. A, silhouette of leaf of C. cordylocarpa ( $\times$  ca. 1/2); B, meiotic chromosomes of C. cordylocarpa.

Bidens cordylocarpa (A. Gray) Crawford, comb. nov. Coreopsis cordylocarpa A. Grav, Proc. Amer. Acad. Arts 22:428, 1887, Fruticose, 0.5-2m tall, stems several from the base, red, hispid or with appressed hairs, becoming glabrous toward the base; leaves opposite, 10-20 cm long (including petiole), deltoid in general outline, pinnately divided, appressedly-pubescent on both surfaces; heads cymosely disposed, mostly 3-12 aggregated, peduncles 2–15 cm long, becoming densely pubescent near the involucre; heads 4-9 cm wide at anthesis; outer involucral bracts 6–10, lanceolate to narrowly so, hispid, 3–9 mm long; inner involucral bracts 8-12, lanceolate to narrowly ovate, hispid, 4-8 mm long; chaff narrowly lanceolate to linear, glabrous or sparsely hispid, 5-8 mm long at anthesis; ray florets 8-16, sometimes in a double whorl, neutral, ligule oblong to oblanceolate or linear, 0.6-4 cm long, 0.4-1.2 cm wide, entire or shallowly notched at the apex; disk florets 20-60, stigma hispid, shortly caudate; achenes club-shaped, essentially terete in cross section, glabrous, weakly striate, wingless, exaristate, and topped by a bald disk.

Representative specimens: MEXICO. Jalisco, bank of stream, 5200 ft. Sierra de San Estéban, *Barnes & Land 155* (F); banks of Río Blanco near Guadalajara, 5000 ft., *Pringle 11506* (F, MICH, MO, MSC, US); gravel along small stream 15 road mi N of Guadalajara, on road to San Cristóbal de la Barranca, 5100 ft., *Cronquist 9817* (F, MICH, MO, MSC, NY, TEX, US); in boulders and sand of stream bed leading into the barranca of the Río Blanco, ca. 8 mi N of Guadalajara, *Mel*-

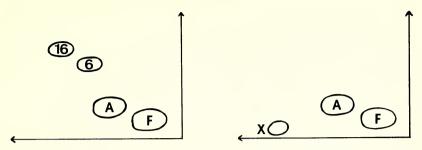


FIG. 5. Drawings of two dimensional chromatographic patterns of flavonoids in *C. cordylocarpa* (horizontal axis = tertiary butyl alcohol run; vertical axis = 15% acetic acid run); left, profile of leaves; right, profile of floral tissues.

chert, Sorensen, & Crawford 6354 (IA, RM); among boulders of swift stream, ca. 3 mi W of Cuaulta along road to Los Volcanes and Puerto Vallarta, Melchert, Sorensen, & Crawford 6371 (IA, RM); among boulders of rocky stream bed 12.5–13 mi N of Zapopan, along dirt road to San Cristóbal de la Barranca, Melchert, Sorensen, & Crawford 6347 A-B (IA, RM); Hwy. 41, 7–8 mi N of Guadalajara, Carman 68-60 (IA, RM).

#### ACKNOWLEDGEMENTS

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#### NOTES AND NEWS

NOTES ON THE FLORA OF THE PACIFIC NORTHWEST.—Extensive collections from Pend Oreille Co., Washington, were made by the author in connection with a floristic study (Layser, E. F. A floristic study of Pend Oreille County, Washington. M.S. thesis, State Univ. New York, College Forestry. 1969). Among the collections, certain ones seem worth special note.

Berteroa incana (L.) DC., a weedy European crucifer, was collected along the roadside in the northern part of Pend Oreille Co. (Layser 1175, WS) and previously